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THE HAUSTORIA OF THE ERYSIPHEÆ.

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(WITH PLATES XI AND XII)

HISTORICAL RÉSUMÉ.

THE early literature on the Erysipheæ is of historical interest chiefly. Until the middle of this century the need of exhaustive observations of the phenomena exhibited by plants had not been felt by botanists generally. The study of plant structure as a means of revealing functions had not been established as opposed to the study of structure as a means of revealing relationships in plants. Hence, though the destruction of crops by parasites had been known since the time of the Greeks and Romans, the idea of parasitism had as yet a vague and erroneous definition. By the middle of the century, however, it seems to have been quite generally accepted that parasitic fungi are nourished in some way by their mycelium, "by imbibing juices impregnated with the peculiar principle of the matrix on which they grow" (28). Thus, though this statement was extremely vague, it looked in the right direction. The mildew of the peach, rose, etc., had long been included in lists of fungi by systematists, yet the study of the structure of the fungus had proceeded only to figuring pieces of hyphæ with their conidiophores and conidia. There was little information at hand, therefore, to assist in understanding the nature of a widespread grape disease soon to be produced by one of these Erysipheæ.

Investigation received a stimulus from economic considerations, when, in 1845, a destructive grape disease appeared at Margate, England. Tucker, a gardener there, was led to a study of a mildew on the vines, because the spread of the fungus kept pace with the symptoms of disease. He published the results of his study in the *Gardeners' Journal* of 1847. The mycelium is described as ramifying through the intercellular spaces of the host-leaves and as sending reproductive branches to the exterior through the stomata. Tucker believed the fungus was a parasite on the vines. He was aided in his investigations by Berkeley (3) who figured and described the fungus as a new species (*Oidium Tuckeri*). Berkeley repeatedly referred (4) to the hemi-endophytic habit of the fungus, and as late as 1886 Worthington G. Smith (30) affirmed that the mycelium may sometimes be found on the interior of the leaves. If Berkeley's observations are correct, *Oidium Tuckeri* would seem to be related to Phyllactinia (p. 175) and to be distinct from the American *Uncinula spiralis*.

From England the disease spread to France and appeared at Versailles in 1848 (9). By 1851 it had spread all over Europe, causing much distress in vine-producing districts. Losses to owners of vineyards were so great that in France and Italy commissions were appointed to investigate the disease. Independently, Von Mohl (33) set for himself the task of determining whether the fungus caused or followed the disease, since Robineau (25) had ascribed the disease to an insect. Von Mohl did not record his methods of investigation, except to state that he examined the interior of the fruit under a microscope (33, p. 14). In this connection he noted the cracking of the larger grapes when covered with fungus. His search in the interior tissues of the fruit did not reveal the presence of the fungus there, so he concluded that the fungus produced its effects through the epidermal cells. Upon the epidermal cells of the young leaves, flowers, and fruit were to be seen, as soon as the fungus became visible, brown spots, which spread as the fungus spread. Young fruit upon which the fungus appeared did not

reach maturity, but withered and died. He concluded that the fungus was a true parasite on the vines and produced the disease. In 1853 Von Mohl (**34**) attempted a serious study of the weather conditions under which the fungus was able to accomplish such destruction. He examined the infested leaves more carefully and learned that the brown spots on the leaves appeared wherever the fungus was attached. At the brown spots the hyphæ produced irregular outgrowths for attachment, which he called *Haftorgane*. He strongly objected to the belief of Amici (**1**) of the Italian commission, who reported that the appearance of mildew on the vine was a proof of disease in the plant rather than the cause of the disease. This was the opinion also of Léveillé (**19**). Von Mohl cited in support of his proposition that the mildew produced the disease, the opinions of Visiani and Zanardini, of the Venetian commission. Zanardini (**38**) had observed in 1851 the protrusions by which the fungus appeared to attach itself, and called them *fulcra*. Von Mohl credits Zanardini, therefore, with the discovery of these *Haftorgane*. According to Von Mohl (**34**, p. 594) Visiani believed that he had found root-like organs arising from these *Haftorgane* and penetrating into the epidermal cells of the grape leaves. But Von Mohl did not accept Visiani's observations as correct, for he had seen only the surface organs of attachment and supposed they acted as suckers for imbibing nutriment. It seems probable, then, that Visiani was the first to see what we now call haustoria, and in his comparison of them to the roots of higher plants, he was the first to obtain a conception of those functions by which the parasite nourishes itself. Thus, as the structure of the fungus became better known, the general notion of parasitism took more definite shape.

Berkeley (**6**) believed with Von Mohl that the fungus caused the disease, and that the surface organs of attachment, which Frank (**14**, p. 556) has so well named appressoria, were suckers which drew up the juices of the host plant. Indeed, Berkeley (**5**) and subsequently other English writers applied the term fulcra to the appendages of the perithecia, and Berkeley at

least believed that they thereby absorbed nutriment from the leaves (2). Later, however, he wrote, "It is very doubtful whether they extract anything from the matrix" (7).

De Bary (12) seems to have been the first to work out the structure of the haustoria. His contributions to the subject are by far the most important and embrace the most of what is known of these organs. He applied the name haustoria, however, both to the exterior organs of attachment (Von Mohl's *Haftorgane*) and to the absorbing organs of the fungus, which he found within the epidermal cells of the host leaves. Frank (14, p. 556), at a later date, named the *Haftorgane* appressoria, reserving the term haustoria for the real organs of absorption. Of haustoria De Bary distinguished three sorts (12, p. 26): (*a*) *haustoria exappendiculata*, found in Sphærotheca, Podosphæra, and such forms of Erysiphe as have two-spored asci, the haustoria originating directly from mycelial filaments, which show little or no contortion at the points where the haustoria arise; (*b*) *haustoria appendiculata*, the haustorium arising from a somewhat hemispherical protrusion of the hypha, *i. e.*, from an appressorium; (*c*) *haustoria lobulata*, or lobed appressoria. In his Comparative Morphology (11) De Bary defined haustoria as "special organs of attachment and suction," arising in the Erysipheæ, from septate mycelial hyphæ. At this point the hypha is firmly attached to the epidermis of the host and sends the very minute haustorial branch directly through the outer wall of the host cell. Within the cell the branch enlarges into "an ellipsoidal or somewhat elongated vesicle filled with protoplasm, which in *Erysiphe graminis* is branched in a peculiar manner." The description in his Beiträge (12) is more detailed. Up to the point where the haustorial branch within the cell enlarges to form the body of the absorbing organ, the slender branch is apparently thick walled. This is not actually the case, for up to that point it is surrounded by a tube-like offset from the epidermal wall which the haustorium pierces. In cross secton, the neck of the haustorium would, therefore, be represented by a small circle, surrounded by a broad shining ring from the

host-cell wall. At the point where the vesicle begins, this collar suddenly becomes thin and becomes continuous with the outer contour of the vesicle. The diameter of a mature haustorium proved to be approximately equal to the diameter of the mycelial thread. When the vesicle is young, De Bary says it has a very delicate wall, but an old haustorium has a colorless double-contoured wall within which is minutely granular protoplasm, either homogeneous throughout, or consisting of a dense ball in the middle surrounded by transparent granular protoplasm. In cases where the haustorium is extremely old the contents consist of shining, oily masses. Not infrequently, if the epidermis to be penetrated is thick or if the penetrating tube ceases to grow, he observed that the haustorium does not enlarge into a vesicle, but is surrounded by a cone-shaped or knob-shaped protrusion of epidermal wall projecting into the cell. De Bary also examined grapes and grape leaves infested with *Oidium Tuckeri*. He seldom found the haustoria normal in the fruit of the grape, but usually he found only the penetrating tubes surrounded by the cone-shaped ingrowths of the browned host-cell wall extending into the epidermal cells, as just described. Even in cells containing mature haustoria he found no great disturbance wrought in the cell wall, the cell protoplasm, or fluid contents. He frequently observed, however, in otherwise normal cells, that the haustoria are surrounded by thick, irregular masses of protoplasm belonging to the host-cells, which ammonia or KOH would remove sufficiently so as to allow the true haustoria to be seen. Gradually, the browning observed on the exterior of the epidermis spread to the contents of the infested cells, but he did not observe such extreme browning as was described by Von Mohl. It is to be noticed that all of De Bary's studies of the haustoria were made from the surface of the leaves.

Büsgen (10) refers to haustoria in reporting his observations on the effects of chemotropic and other stimuli upon the germinating tubes of fungi. He germinated conidia of *Erysiphe communis* from leaves of *Polygonum aviculare* on slides under cover glasses. In some cases he used sections of infested leaves

in weak nutrient solutions. He was able to obtain appressoria-like organs where the filaments came in contact with the cover glass, but the germinating tubes had a very limited growth and soon perished. His results with the leaves indicated chemotropic reactions in the filaments. As a whole his experiments on the Erysipheæ were not directly successful and they bear little relation to the haustoria themselves. Reference will be made to this paper again in discussing Phyllactinia.

Galloway (**15**) devotes a short paragraph to haustoria in an account of his observations on the development of *Uncinula spiralis* and figures them in much the same way as De Bary. Harper very briefly describes the haustoria of *Sphaerotheca Castagnei*, and he gives but a single figure (**17**). Palla (**24**) has recently investigated Phyllactinia on Berberis and Corylus, but he concerns himself rather with the habit and structure of the mycelium than of the haustoria. For the minute points of structure his methods are inadequate, as he himself intimates (p. 70).

It will be seen from this brief résumé of the literature that the minute structure and especially the development of the haustoria are almost entirely unknown.

METHODS.

Four fixing solutions were experimented with and their respective merits were compared: Flemming's fluid (stronger solution), Merkel's solution, chrom-acetic acid (0.7 per cent. of the former and 0.3 per cent. of the latter), and a saturated solution of mercuric chlorid in 1 per cent. acetic acid. Flemming's fluid proved the most reliable and satisfactory. In those cases in which it blackened the tissues, the sections, after having been attached to the slides, were bleached for twenty-four hours in hydrogen peroxid before staining. Merkel's solution was frequently satisfactory, but it proved hardly as reliable as Flemming's. The other two solutions were far less useful. The sections of leaves were cut 6–7 μ in thickness and were fixed to the slides by the well-known albumen and distilled water methods. They were cleared in clove oil and mounted in Canada balsam.

Some difficulty was experienced in sectioning those leaves which contain large crystals of calcium oxalate, as Corylus and Xanthoxylum, because these crystals frequently caused the destruction of the ribbons. For that reason, the side of the leaf bearing the fungus was usually turned toward the knife in cutting, unless the fungus was amphigenous.

All staining was done on the slide. Wisselingh (**37**) finds in fresh material that chitin is present in many places as one constituent of fungus cellulose. In the Erysipheæ, chitin appears in the perithecia, appendages, mycelium, conidia, and conidiophores. He does not mention haustoria in this connection and finds no characteristic stain for walls containing both cellulose and chitin ; congo-red, however, in neutral and ammoniacal solutions, stains pure cellulose intensely, while if chitin is present also, the color is not dark, unless the chitin be transformed to mycosin.[1] Mangin (**21**) found pectin quite generally present in fungus cellulose and employed ruthenium-red as a characteristic stain. Wisselingh (**37**, p. 632) was able to remove the pectin from cell walls containing it. Mangin did not investigate this question and Wisselingh denies the presence of pectin in the Erysipheæ. My own results confirm this view. With ruthenium-red the fungus would not stain at all, though the middle lamellæ of the cells of the host-leaf gave the pectin reaction. The most characteristic staining obtained in the fungus, in the use of congo-red (neutral and alkaline solutions) and methylene-blue (neutral and acid solutions), was an intense staining of the ascus wall in the perithecium. Wisselingh does not mention this as a part containing chitin, and, since the remainder of the fungus seemed to give the chitin reaction by refusing to stain deeply with congo-red, it is possible that the ascus wall is composed of pure cellulose.

The walls of the haustoria do not show any reactions to stains that would indicate that they contain any different material from the remainder of the fungus. Flemming's well-known triple stain, safranin, gentian-violet, and orange-G, proved

[1] He used a modification of Gilson's method (**16**).

very satisfactory. By this stain the nucleoli of the fungus and host are stained red, the chromatin blue, and the ordinary protoplasm orange. Fungus nuclei in the process of division take the violet stain readily, but in a resting condition they possess chromatin in very minute masses. The most of the nuclei shown in the accompanying figures had to be under-stained with violet in order to prevent the over-staining of the host-tissues. The nuclei are represented as containing conspicuous chromatin granules, because in properly stained nuclei the granules are differentiated.

ERYSIPHE COMMUNIS.

Erysiphe communis proved very favorable for study, and since Erysiphe, Sphærotheca, Microsphæra, and Podosphæra appear to agree closely in respect to their haustoria, the fungus on *Geranium maculatum* will be described as a basis for comparison. *Fig. 8* represents such a case as De Bary described. The hypha from which the haustorium originated is not shown, as the mycelium is very easily so far separated from the leaves in the process of fixing and imbedding as to destroy the connection between haustoria and hyphæ, a fact upon which De Bary (**12**, p. 27) and Harper (**17**) comment. The absorbing organ is seen to consist of a slender, proximal portion, the neck, penetrating the epidermal wall of the cell, within which it enlarges into a vesicular, distal portion with a thin wall. On the interior, the vesicle is filled with a delicate, spongy protoplasm differing in no visible particulars from the protoplasm in the mycelium. A mature haustorium always contains one normal nucleus, a fact to which De Bary does not refer. He probably saw it as the "thick ball" surrounded by transparent protoplasm, to which reference has been made. Harper (**17**) for the first time mentions the nucleus in his paper. *Fig. 11* represents the only case I have seen of a haustorium with two nuclei. This haustorium is also septate and such septa occasionally present themselves (*fig. 13*). *Figs. 8, 19*, etc. show in longitudinal section that the ingrowth from the inner surface of the wall of the host-cell

surrounds and accompanies the neck for a distance, as De Bary reported. It seldom extends down to the body of the haustorium, however, and sometimes is absent altogether (*fig. 16*). The collar from the cell-wall is somewhat different from the wall from which it takes its origin, and usually stains little, while the remaining portion stains with safranin intensely. The outer surface of the neck adheres closely to this collar, the outer boundary of which is represented by the middle circle in *fig. 17*.

But *fig. 8* does not represent the usual conditions in one particular. Almost always a mature haustorium is surrounded by a thick, sheath-like layer which, De Bary says, belongs to the protoplasm of the host-cell. It is clear that, by the use of hand-cut sections of fresh or alcoholic material and by a study of the haustoria from the leaf surface, he was not able to make out the structure of this sheath fully. De Bary describes it as an irregular mass; but it is not extremely irregular and is bounded by a very thin membrane of about the delicacy of the plasmic membrane of the host-cell. In *fig. 9*, for example, the middle cell contains an optical section of a haustorium. Within the center lies the relatively large nucleus with its red-staining nucleolus and fine chromatin granules. Surrounding the nucleus is the spongy protoplasm staining orange. The haustorium-wall surrounds this protoplasm. Exterior to the haustorium are the contents of the sheath described by De Bary as belonging to the protoplasm of the cell, while bounding the whole, in contact with the contents of the host-cell, is the delicate limiting membrane of the sheath itself.

The substance of this sheath looks like protoplasm at first glance and stains with orange, but it is not vacuolated like protoplasm. It consists of a dense, homogeneous, finely-granular mass, most frequently gathered into lumps of varying outline, which appear very slightly granular (*figs. 9, 11, 16*). Rosen (**26,** p. 258) observed in *Puccinia asarina*, growing in the intercellular spaces of Asarum, that the branched haustorium was connected with the nucleus of the host-cell in the majority of cases, and either adhered closely to it or entered it with disorganizing

effect. Harper (**17**, p. 664), disagreeing with De Bary, thinks that among the Erysipheæ this peculiar sheath is, as in Puccinia, the disorganizing nucleus of host-cell. The delicate bounding membrane would, therefore, be the nuclear membrane.

I have not found evidence of such a relation between haustorium and host nucleus in the Erysipheæ. In the examination of many sections in searching for stages in the development of the haustoria, it becomes more and more clear that the host-nuclei and haustoria are indifferent to each other. The sheaths are present around the haustoria even if there are several infesting one cell, as frequently happens. In such cases, if the cell is not so full of haustoria as to obscure the cell contents or crowd the nucleus, that body can be seen in a more or less normal condition. In the cells of hairs which are large enough to contain several haustoria easily, the sheath of each can be seen, while the host-nuclei occupy distant positions in the cells. The sheaths are to be accounted for in another way, as will be shown subsequently.

In most of the figures the host-nuclei lie at a greater or less distance from the haustoria. When, as in *fig. 16*, the connection between the two is close, the nuclei are more or less disorganized. But the cases figured make the proportion seem much too large to be in accordance with the facts. The sheath is not usually bounded by a tensely stretched membrane (*fig. 7* represents a special case), but by a membrane having a more or less irregular outline in section, beginning where the cellulose collar stops. The orange-staining contents are usually present in more or less abundance, except in *Erysiphe graminis* on Poa, where only the sheath is present and is sometimes inconspicuous.

The outer walls of the epidermal cells have an affinity for the safranin stain, but the collar about the haustorium neck stains much more delicately with it. In *fig. 10*, shown because the cell was plasmolyzed, the collar is distinct from other parts and appears under the microscope very slightly stained with safranin. Again (as in *figs. 16, 21*, and frequently in other figures), the reaction is such that the neck of the penetrating organ is

distinctly visible and its slenderness appreciable; whereas, if it were not for this circumstance, the size of the tube would frequently be exaggerated.

Nordhausen, in experimenting with Botrytis, Penicillium, and Mucor (**23**, p. 38), observed a marked browning of the epidermal cells in contact with the germinating spores, which always preceded penetration. He thinks (p. 7) that the phenomena of browning and subsequent death of the cells are due to the production of some poison during the germination of the spores. Von Mohl (**34**, p. 592), De Bary, Frank (**14**, p. 556), and others mention this browning in the Erysipheæ. In the stained sections this brown color is not distinguishable, though the cell wall around the point of penetration is more or less altered and dissolved. Seen from the outer surface of Poa (*Erysiphe communis*) and Eupatorium (*Erysiphe Cichoracearum*), there is an area surrounding the point of penetration which is entirely colorless, clear, and shining. The remaining portions of the epidermal wall stain with safranin. The outer surface of the colorless area is usually depressed also, the depression being deepest at the point of penetration, as though a part of the cellulose had been dissolved away. This dissolution of the cellulose suggests that the Erysipheæ probably produce an enzyme suited to this work.

In the sections of Geranium leaves the safranin stain frequently makes evident the beginning of penetration. The first intimation of the process is a deep staining of the inner surface of the outer wall of the cell immediately under the point where the hypha comes in contact with the epidermis (*fig. 1*). This staining, however, is to be seen occasionally at the cross-walls of the epidermis, where haustoria have never been produced. It is possible that it is this deeply staining spot which shows the brown color in fresh material.

The next step to be observed in the development of the haustoria (*fig. 2*) is the thickening of the epidermal wall toward the interior over an area coinciding roughly with the clear space already mentioned (*fig. 17*). Nordhausen (**23**, p. 17) observed

a slight swelling of this kind with Botrytis on Tradescantia, Mnium, etc., but Ward (**35**) observed with the same fungus (apparently), which produced the lily disease he described, such an extraordinary swelling of the walls of the host that a large part of the lumen of the cells was filled. Nordhausen believes that Botrytis lives on the protoplasm of the poisoned cells chiefly. Ward is of the opinion that the fungus causing the lily-disease lives also on the gelatinized walls. Erysiphe appears to produce not so much a swelling of the wall of the cell as the addition of new material to its inner surface, for the collar of the haustorium, formed from a part of this thickening, is dense and remains as a permanent structure in the cell.

At the same time that the wall of the cell is thickening, growth of the penetrating tube is proceeding. Its distal end enters the wall, and, just at the point where the reddening of the wall originally appeared (*fig. 2*) a very slight enlargement of the tube occurs, accompanied still by the reddening on each side (in longitudinal section). But this effort does not bring the point of the tube into the lumen of the cell, for the thickening of the wall keeps pace for a time with the growth of the tube. This tube is extremely minute. For Botrytis Nordhausen (**23,** p. 39) found it to be one fourth of the diameter of the ordinary hyphae. Miyoshi (**22**)[2] has shown that the membrane to be penetrated affects the size of the tubes. Thus, when collodion was used for a membrane, the tubes actually increased in diameter, while with an onion skin there was no change in the size of the tube. Ward's observations on this point agree with Nordhausen's. In all of the Erysipheæ the tube is much smaller, as the figures show, and it is interesting to note that the nucleus of the absorbing organ must in some manner make its way through this minute passage.

The tube (*fig. 3*) continues its growth through the increasing or (as Ward thinks for Botrytis) swelling cellulose, a part of which remains permanently encircling the neck of the mature

[2] Die Durchborung von Membranen durch Pilzfäden. Jahrb. f. wiss. Bot. 30: 280. 1895.

haustorium as the collar so frequently mentioned. De Bary (12 p. 26) probably saw something of this phenomenon in the form of the knob-shaped papillæ in the cells as mentioned above. He saw the structures in cells with thick epidermis and in what he thought were no longer growing haustoria. Because of the closely connected series of stages which it is possible to secure, it is evident that these are not old, disintegrating haustoria. Rather they represent the early stages of penetration. At any rate, if they represent dead haustoria, they reveal the outlines of the cellulose parts none the less, and so tell the story of development. Just what significance there is in this thickening of the host wall cannot be conclusively determined without experiment. The wall increases not only in bulk, as Ward found, but in quantity, as has been mentioned. That there soon *does* appear to be a disintegration of a portion of this ingrowth, will be seen presently. There are not data at hand to determine whether the penetrating tube, by means of some chemical substance, excites the cell protoplasm to unusual activity in the production of cellulose over t he region of penetration, or whether microscopically small needles would cause such a production mechanically. Again, it is possible that the stimulating agent is the atmosphere acting through the wall at the point made thin by the work of the fungus. There would then be the same reason for this local thickening as there was for the original production of a thick outer wall in the first place.

Soon the tube, growing with increasing rapidity, overtakes the cell in what may be its efforts to protect itself from injury by the fungus. Whatever may have been the stimulus which produced it, the thickening of the wall ceases after a considerable ingrowth, U-shaped in outline, has been formed. The first appearance of disintegration is now to be seen in it. The distal end no longer has the appearance of cellulose such as is found in the cell wall and in the clear basal portion. Some change has been wrought in it which has altered its appearance and its reaction to stains. This end (*fig. 4*) may now be stained slightly orange, and it is slightly and minutely granular. No

sharp boundary separates the proximal from the distal end. The granular character of the latter gradually lessens toward the dense proximal end, which stains very slightly with safranin. The still slender penetrating tube is to be seen piercing this basal portion and extending into the now disintegrating distal extremity. It is to be observed that the plasmic membrane of the cell, by means of which the ingrowth has been formed, passes up the side and over the end of that structure. The gradual disintegration of this ingrowth of cellulose thus forms the early stages from which the true structure of the haustorium sheath can be understood.

The growth of the haustorium now continues with accelerated speed. It will be seen in *fig.* 5 that the penetrating tube has taken a straight course through the cell wall and the basal portion of the papilla, for a distance approximately equal to the length of the collar in a mature haustorium. From that point on to the point where its growth was checked by the fixing fluid, the tube has pursued a rather tortuous way or else the distal surface of ingrowth has offered some resistance to the progress of the tube. The end of the tube has begun to enlarge into the vesicle which forms the body of the mature haustorium. The nucleus has not yet started in. The whole distal portion of the cellulose surrounding the end of the tube is now distinctly granular and takes a deep orange stain. The staining of the wall on the line of its original inner surface is no longer visible. The hypha from which this tube originated is sufficiently enlarged and flattened to fall within De Bary's class of *haustoria appendiculata*, peculiar to forms of this genus possessing two-spored asci. *Erysiphe communis*, however, would be a species without appressoria, according to this classification.[3]

[3] It seems questionable whether much dependence can be placed upon forms of appressoria in systematic determinations, such as the separation of *E. Galeopsidis* from *E. Cichoracearum* (Ellis and Everhart, N. A. Pyrenomycetes 14. 1892). The former is supposed to have lobed appressoria. The varieties of *E. Galeopsidis* used in this investigation do not appear to show more conspicuous appressoria than *fig.* 5 represents. Between the appressorium and the epidermal wall is drawn in outline a cushion-like structure which stained red. This was the only case of the kind observed, and its significance is unknown. In *fig.* 5 again the nucleus of the host is on the basal wall of the cell.

In *fig.* 6 the absorbing organ is in a thick-walled epidermal cell in the region of the midrib of the leaf. The vesicle has continued to enlarge until it has reached the form, but not the size of a mature haustorium. It would never have possessed the usual sheath, because by pressure or fermentative action, or by both, it has escaped from its enclosing cellulose, which has approximately the same structure as the disintegrating papilla shown in *fig.* 5. The collar at its base, therefore, consists of the entire ingrowth and not of its basal portion only, as is common in most mature haustoria. It does not as yet contain a nucleus. Outside of the cell is shown a piece of mycelium containing numerous safranin-staining bodies which are probably food material of some sort.

In *fig.* 7 is shown a nucleus making what appears to be an unsuccessful attempt to enter the haustorium. The hypha has been broken away so far that none of the pieces lying above the cell could be identified surely as belonging to the haustorium. The nucleus seems to find some difficulty in entering, either because of its size or, possibly, because the darkly staining material lying in front of it offers some resistance to its progress. The passage of the nuclei into the haustoria is naturally most difficult to see. This unsatisfactory example is the only case observed. The haustorium must attain nearly its full size, therefore, before it is provided with a nucleus. Up to this time it contains only highly vacuolated protoplasm. It is difficult to find cases which show nuclei in position to pass down the penetrating tubes into the haustoria. The few cases observed have been figured, but it cannot be said conclusively that these are the nuclei which were ultimately to find their way into the haustoria. The nuclei of the hyphæ are sometimes greatly elongated and narrowed (*fig.* 14). It must be in such a form that they make their way into the tube.[4] *Fig.* 7 is instructive in another respect. The sheath of the haustorium contained, when the material was fixed, nothing but fluid. The contents have been entirely consumed. But the bounding membrane is

[4]See also figures by Harper, 17, p. 663.

stretched and turgid, and none of the solid contents of the host-cell approaches nearer to the haustorium than the outside of this *sheath-membrane*. That this membrane is so tensely stretched shows that it was filled with a liquid when the material was fixed. It, therefore, possessed osmotic qualities similar to the plasmic membrane of the host-cell. It has been mentioned that the plasmic membrane of the cell, by the activity of which the cellulose papilla was produced, extends over the papilla (*fig. 4*). In *fig. 7* this plasmic membrane is stretched and enlarged extraordinarily until its origin would not be easily recognized.

From what has been said on the development of the haustoria, it is easy to understand the nature of the sheath around the haustorium, with its bounding membrane. That is, it does not belong to the protoplasm of the cell as De Bary supposed, nor is it the host-nucleus as Rosen observed in *Puccinia;* but the contents of the sheath consist of disintegrated cellulose from the distal end of the cellulose ingrowth through which the haustorium has made its way. The bounding membrane of the sheath, on the other hand, is the plasmic membrane of the host-cell stretched and greatly enlarged by the osmotic forces involved. In so far only does the sheath belong to the protoplasm of the cell, and not at all in the sense which De Bary had in mind. There is abundant evidence from various sources to support the view just stated. Marshall Ward (**35**, p. 356) has shown that Botrytis produces a swelling and gelatinization of the cell-walls of the lily leaves. Ward found that the fungus was able to live on this disintegrated cellulose. It is well known that certain fungi produce ferments which are able to digest cellulose. Ward found such a ferment in Botrytis (pp. 343–346). So also Beyerinck (**8**) found that *Coryneum Beyerinckii* makes use of a dissolving ferment. Nordhausen (*l. c.*, p. 38) has shown that Botrytis, Penicillium, and Mucor can enter a cell-wall and grow through it parallel to its surface for comparatively long distances. Indeed, this power of disintegrating cellulose is probably generally possessed by fungi, parasitic and saprophytic. The Erysipheæ probably have such a ferment. The partial

dissolution of the epidermal wall of the host about the point of penetration has been mentioned. The penetrating tube makes its way through a long ingrowth of cellulose before it expands into a mature haustorium. The mature organ shows no unmodified host-cellulose surrounding it except the collar around the neck of the haustorium. The distal portion of the ingrowth partially disappears in the development of the penetrating tube. During the disappearance, it undergoes changes which materially alter its microscopic appearance as concerns structure and its capacity of reacting to stains. It becomes decidedly granular and takes an orange stain, whereas, in the beginning, the entire ingrowth was of the same consistency as the collar, and took the safranin stain slightly. By the time the penetrating tubes begin to enlarge, these changes are conspicuous. The tube may finally break through the cellulose and develop without any sheath. But usually some of the cellulose remains as the granular masses of the sheath. It will be shown subsequently that, under certain conditions, the haustoria of *Uncinula Salicis* have no sheaths, and the sheaths of *Erysiphe graminis* on Poa do not show any contents in the older stages nor are the sheaths always present.

The extremely minute size of the penetrating tube has been mentioned. The amount of fermentative action, of which the young haustorium is capable at first, is, therefore, only sufficient to provide for the onward growth of the tube. The cell succeeds in laying down cellulose ahead of it for a time. The circle of the fermentative effect has not at first a long radius. The distal end of the ingrowth is the first to show signs of dissolution. It becomes granular there. As the absorbing organ attains larger growth the digestive powers of the fungus become more effective, and that part of the collar coming within the sphere of influence of the ferment is gradually attacked and partly dissolved.

It is interesting to find that the membrane by which the sheath is bounded in a majority of cases is contributed from the plasmic membrane of the host-cell. The plasmic membrane is

not ruptured by the ingrowth of cellulose. Rather the ingrowth is caused by the activity of the membrane, the area of which enlarges with the thickening of the wall. The plasmic membrane is still recognizable when the haustorium begins to enlarge at its distal end. It still maintains its osmotic properties at the stage represented by *fig.* 7. In most cases it rémains bounding the masses of disintegrated cellulose which constitute the contents of the sheath. *Fig.* 7 represents the membrane stretched and firm from the osmotic forces at work in the nutrition of the fungus. At a later stage it usually suffers injury and sometimes dissolution.

This fungus is not capable of producing the extraordinary dissolution of cellulose which Ward found in the case of Botrytis. Its supply of enzymic material seems to be limited. The sheaths are, therefore, usually present, though on rare occasions they are not, as in *figs.* 8 and 12. Certain haustoria of *Uncinula Salicis* do not possess sheaths and, as will be seen, this species has a greater digestive capacity. Nordhausen (**23**, p. 23) found in infecting leaves with spores of Botrytis that heavy dews so weakened the enzyme of the fungus that penetration was impossible. It seems probable that, when the cell-sap of Geranium begins to be absorbed by the young haustoria of Erysiphe, the enzymic material is weakened or largely reabsorbed. Thus it can be seen how the sheaths with their bounding membranes are possible and how, as De Bary observed long ago, the host-cells may escape with so little injury. From the time when the haustoria begin to absorb actively, the further digestion of the cellulose papillæ largely ceases, and though their bounding membranes collapse, they are usually not completely digested, being protected by the cell-sap. In *fig.* 8 the sheath is all digested down to the collar. In *fig.* 19 the haustoria succeeded in penetrating the wall of the hair, of which the cell forms a part, before the plasmic membrane could build up papillæ to impede the progress of the tubes. The digestive capacity of the haustoria is not entirely constant, so that we find sheaths in various stages of disorganization.

ERYSIPHE GRAMINIS AND OTHER SPECIES.

The absorbing organs of *Erysiphe graminis* on *Poa pratensis* deserve description. It has long been known that the haustoria of this species are "branched in a peculiar manner." This fungus grows very luxuriantly on the grasses and fills the epidermal cells full of large branched haustoria. A shining, colorless area occurs around the penetrating tube (*fig. 17*) which is slightly larger than in *E. communis*. The colorless area is slightly depressed, as already mentioned. The collars of cellulose are relatively thick also. *Fig. 19* represents nearly the maximum thickness for the collars in Poa. The body portion of the absorbing organ is always approximately cylindrical or ellipsoidal, with finger-like projections growing out from the ends or sometimes from only one end (*figs. 19, 21*). A large nucleus lies near the middle, and in either end, in mature examples, there is a large vacuole. The branches are also vacuolated. The body is not always symmetrical with respect to the neck (*fig. 20*) but the neck may be near one end from which, even then, branches may arise. The development of the haustoria of this species was not followed, but in none of the mature examples have the sheaths possessed any granular contents as found in *E. communis* and elsewhere. The sheath-membranes are usually present, though they exhibit great irregularities. Sometimes they are not to be seen except for a short distance; sometimes they completely surround the haustoria, being discernible even down between the branches, At other times the branches penetrate the sheath. The protoplasmic contents of the epidermal cells are usually scanty. It seems impossible that the cell nucleus should escape destruction when the haustoria are thus provided with long branches. But the same indifference to the nucleus exists here as in *E. communis*. Even when a cell contains several haustoria, the nucleus is usually unmolested, and is often as normal in appearance as in uninfested cells. This form of absorbing organ may be looked upon as the result of a special effort of this species to obtain abundant food. The heavy growth of mycelium and the immense number of conidia produced by this species

show the success it has attained. Many of the appressoria of this species agree with De Bary's *haustoria appendiculata*. *Fig. 19*, being a side view, shows only one outline of the organ of attachment. Long infested blades of Poa show signs of injury from the fungus. "In California it has been destructive to wheat" (**13**).[5] Certainly these haustoria give the impression of activity not gained from other Erysipheæ.

The habit or structure of the other species of Erysiphe studied do not differ so much from the account given of *E. communis* that a detailed description is necessary. Basal (and even higher) cells of hairs are especially favorable places for the study of haustoria. *Fig. 16* represents such a cell from a hair of *Eupatorium perfoliatum* infested by *Erysiphe Cichoracearum*. The nucleus of the cell lies between two of the haustoria and seems to be disorganized. The cell is slightly plasmolyzed. The dark globules (stained red in the sections) do not seem to be degeneration products due to the action of the fungus, for the cells in uninfested leaves contain them. They are probably due to the fixing reagents. This plant, it is well known, contains a volatile oil and a resin. It is probable that the globules are related to these substances. It is very common to find such products as these in the autumn, when this material was gathered.

UNCINULA SALICIS.

Uncinula Salicis on *Salix discolor* exhibits peculiarities in its appressoria and haustoria, which apparently have not been reported heretofore. De Bary (**12**, p. 27) mentioned the lobed appressoria (*haustoria lobulata*) of this species, then called *U. adunca*, but he did not report any peculiarities in the haustoria; and Galloway examined *U. spiralis* (*U. necator*). It has frequently been stated by De Bary and others (**12**; **32**; **36**; **20**; **18**; **14**, p. 555; **13**, p. 2; **19**) that the Erysipheæ always confine their absorbing organs to the epidermal cells of the host. Berkeley, it has been mentioned, believed in the hemi-endophytic habit of

[5] The writer has never found it difficult to collect material, with ascospores developed, in August and September.

some of the powdery-mildews, and Palla (**24**, p. 68) has recently shown that Phyllactinia has intercellular hyphæ. *Uncinula Salicis* also offers a striking contradiction to that old conception, but the method by which this fungus reaches the interior tissues of the leaves is different from the one Phyllactinia employs.

Uncinula is amphigenous on the leaves of the willow. Its appressoria are lobed, as De Bary affirmed. The mycelium is entirely external. On the upper surface of the leaves the lobed appressoria give rise to penetrating tubes which enter the epidermal cells. All of these tubes do not develop in the epidermal cells into haustoria. An examination of a cross-section of the willow-leaf shows that the epidermal cells are very abundantly infested with haustoria, but in addition, numerous slender bars can be observed reaching from the outer walls across the epidermal cells to their inner walls. The bars give the cells the appearance of possessing trabeculæ. The haustoria in the cells frequently hide the outer end of these structures, but in following their course to the inner wall, haustoria may sometimes be observed in the palisade cells of the leaf. The bars are the penetrating tubes, or the necks, of these subepidermal haustoria. When the penetrating tubes reach the palisade cells they enlarge into haustoria not unlike the ones described for *E. communis*, possessing the sheaths and nuclei. Several tubes may pierce the outer epidermal wall close together and take different directions across the cells. Of the tubes which penetrate the outer wall of the epidermal cell close together, some may enlarge immediately into haustoria, while others may pass to the palisade cells. This crowding and confusion makes it difficult to discern the true structure. At first glance it looks at times as if the short necks of the epidermal haustoria give rise to one or more branches which develop into haustoria. The deception arises from the crowded position of the organs.

On the under side of the leaf the epidermal cells are likewise penetrated by the slender tubes, some of which immediately enlarge into haustoria, and some of them (a little less than half), penetrate into the mesophyll cells immediately under the

epidermal cells. *Figs. 22–26* all represent cells on the under side of the leaves. The upper epidermal cells of the willow leaves in autumn are so filled with deeply staining products as to make the cells of this epidermis unfavorable for study.

As far as they have been observed the haustoria of *Uncinula Salicis* are not occasionally septate as in *Erysiphe communis*. In the possession of a single nucleus, surrounded by spongy protoplasm, and in its general shape, the haustoria of Uncinula are not peculiar. The subepidermal haustoria are also surrounded by sheaths such as *Erysiphe communis* has. But in the epidermal cells the sheaths are very frequently absent. The explanation of this absence of sheaths lies in the fact that Uncinula is capable of greater enzymic activity than Erysiphe, so that the sheaths are dissolved away. But by the time the tube has penetrated a palisade cell its supply of ferment is either exhausted or weakened (or both), so that the sheaths remain. The typical collar may sometimes be seen around the necks of the haustoria (*fig. 23*), but for the subepidermal haustoria it can be demonstrated at times, from the size and reaction to stains, that the penetrating tube is accompanied entirely across the epidermal cell by the cellulose ingrowth from the outer wall (*figs. 22, 24*).

De Bary does not mention the fact that the lobed appressorium of Uncinula may give rise to several haustoria (*figs. 22, 24*). Other appressoria, as far as they have been observed, produce absorbing organs singly, but on the willow leaf two or more tubes from the same appressorium pierce the epidermis close together. The confusion and crowding of haustoria in the cells is due to this fact, rather than to the fact that the extremely dense mycelium sends many single organs into the leaf. On the other hand, the lobed character of the appressoria seems to be produced by the contortion of the filaments necessitated by the origin of several haustoria in one place. The significance of the term appressoria consists in the fact that the expansions of the filaments which rise to haustoria are commonly closely appressed to the surface. But this habit is not uniform in *Uncinula Salicis*. *Figs. 23, 25*, and *26* show that the appressoria may stand at some

distance from the surface of the host, and the penetrating tubes may have a shorter or longer course before they pierce the cells. Whether such appressoria have been forced away from the surface of the leaves by the resistance offered to the elongating penetrating tubes by the epidermis is not known.

It might be supposed that, where an intercellular space occurs between two subepidermal cells, the penetrating tube might find its way into the interior of the leaf. The haustoria of Uncinula, however, have not been found deeper than the subepidermal cells.

PHYLLACTINIA.

Palla (**24**, p. 68) has recently reported that Phyllactinia[6] has the very interesting habit of sending nutrient hyphæ through the stomata into the intercellular spaces of the infested leaves. Haustoria are thus constructed entirely on the interior of the leaf[7] and are not found in the epidermal cells as in Erysiphe. Because of this unusual habit Palla suggests the separation of the powdery-mildews into two families, the Erysipheæ and the Phyllactineæ. Upon the ground of certain differences, chiefly in the appendages of the perithecia, he gives (p. 65) the name *P. Berberidis* to the fungus on Berberis, as contrasted with *P. suffulta* on leaves of *Corylus Avellana*. No effort has been made to compare the material collected for this investigation with Palla's results for systematic purposes, but the writer can confirm most of Palla's observations on the intercellular hyphæ and haustoria. Palla finds that the haustoria on the intercellular hyphæ are found in the mesophyll cells in the two hosts he examined. As shown from a study of several hosts, Corylus,

[6] It seems worthy of mention that, on mature perithecia, the appendages in this genus do not extend parallel to the surface of the leaf, as in the young stages, but obliquely downward. The result is that the perithecia are raised into the air for the length of the appendages. The perithecia, therefore, fall off easily when the leaves are handled. It may be that the appendages serve the fungus as organs of distribution.

[7] Investigators who have thought of the Erysipheæ as purely epiphytic parasites, or as receiving their nutriment entirely from the epidermal cells, have been cited on p. 172. See also **27, 29, 31.**

Fraxinus, Cratægus, and Kornus, haustoria pierce the cells of the loose parenchyma in a minority of cases. Usually the intercellular hyphæ first penetrate the leaf to a region intimately associated with the fibro-vascular bundles (*fig. 31*) before producing absorbing organs. Sometimes haustoria are found in the cells between the bundles and the palisade cells, and sometimes in the palisade cells themselves. Palla observed hyphæ passing into the palisade layer, but he did not find haustoria there. In Xanthoxylum, however, haustoria frequently penetrate the cells of the loose parenchyma (*fig. 27*.) *Fig. 32* shows that the intercellular hyphæ which enter the stomata are either side branches of exterior hyphæ or the ends of original hyphæ. Except when the interior hyphæ arise as side branches immediately over the stomata, it is impossible to determine whether we have to do with an original germinating tube or not, so long as cross sections of leaves are used. If the stoma stands wide open, the initial cell of the penetrating hypha is not narrowed (*figs. 27, 28*), but upon the closing of the stoma the initial cell accommodates itself to the space left to it. It thus is narrow at the middle and becomes larger at the distal end (*figs. 30, 33*). Palla finds that the intercellular hyphæ of *P. suffulta* contain at most three cells (**24**, p. 7), and of *P. Berberidis* two (seldom three) more. The cells of the hyphæ within the leaves examined by me vary with the distance to be traveled before the production of an absorbing organ. Fewer than two cells were not found. The number is typically three to five. The distal cell of the intercellular hyphæ is sometimes extremely long. It is possible that, in tracing the sinuosities of the hyphæ through several sections, septa and nuclei may have been overlooked which would raise the number of cells to more than five. When the hyphæ produce their absorbing organs near the stomata, such hyphæ are noticeably larger than the surface hyphæ. But when the filaments extend a long distance through the intercellular spaces, some or all of the cells are more or less attenuated (particularly the distal cells.) An intercellular hypha may sometimes extend through more than a dozen sections. Haustoria do not appear

at the sides of the hyphæ exclusively, as Palla observed (**24**, p. 70), but frequently from the end, as in *figs. 29, 31*. In all of the hosts mentioned, excepting Xanthoxylum, the haustoria agree in structure with those of *E. communis*. *Fig. 31* shows the haustorium in one of the big parenchyma cells which form a sheath for the bundle. It also arises from the end of a hypha and contains a crystal which is so large as to distort the absorbing organ.

The haustoria of Phyllactinia on *Xanthoxylum Americanum* offer some striking differences in comparison with the ones just described. The intercellular hyphæ have thicker walls and the intercellular appressoria are conspicuous, flattened, sucker-like structures appressed tightly to the cells of the leaf (*fig. 28*). The leaves of Xanthoxylum have a thin layer of loose parenchyma. The bundles are near the lower epidermis. These facts of structure seem to influence the parasite. Its intercellular hyphæ, as a rule, are short and the cells are thick. They are more vigorous in appearance, the walls are less delicate, and they stain more readily than the surface hyphæ. Since the leaves of the host are thin the intercellular hyphæ are shorter than in those hosts where the haustoria are developed far from the stomata. No appressoria were observed on the surface hyphæ, though no special effort was made to find them. The appressoria of the intercellular hyphæ are numerous and conspicuous. Even beyond the point where the haustorium is produced, the hypha may adhere to the cell (*fig. 27*). The chief point of interest, however, is the absence of typical haustoria. In a long search through many sections I was unable to identify surely a single absorbing organ having the typical structure for haustoria of the Erysipheæ. There are numerous absorbing organs such as are represented by *figs. 27* and *29*. The penetrating tubes which pierce the host-cells are as minute as in Erysiphe, but the vesicles within the cells appear to have no protoplasmic contents. They have thick shining walls which, in exactly longitudinal sections, appear to be continuous with the walls of the host-cells. It was supposed at first that they were young haustoria differing

in the details of their development from the haustoria of Erysiphe; but their abundance and the many weeks for which the prickly-ash bush must have been infested when the material was gathered, make it evident that they are modified haustoria. It is to be seen that there is some resemblance between *figs. 27* and *29* and the early stages of penetration represented in *figs. 2* and *3* from Erysiphe. The haustoria of the fungus on Xanthoxylum certainly answer very well to the knob-shaped structures De Bary described, which were probably young haustoria. It is impossible to decide absolutely that they were, however, because of their similarity to *figs. 27* and *29*. Certainly if they represent dead haustoria, as De Bary thought, the vigorous mycelium must be nourished in some other way.

GENERAL CONSIDERATIONS.

The phenomena exhibited by the intercellular hyphæ of Phyllactinia are interesting in connection with what has been ascertained by several investigators in relation to the nutrition of fungi. It has been mentioned that the majority of the intercellular hyphæ in several of the hosts studied (excepting Xanthoxylum) take a more or less direct course to the regions near the bundles. The development by the fungus of the absorbing organs in regions abundantly supplied with available food, such as the parenchyma sheath of the bundles, indicates a selective chemotropism in the fungus. A selective chemotropism was reported by Miyoshi for several fungi in his paper already cited. Phyllactinia thus offers under normal conditions of growth a demonstration of the selective reaction which Miyoshi demonstrated by artificial means. This reaction enables Phyllactinia not only to surround itself with conditions which will insure it an uninterrupted supply of food and water such as is not insured to it while living as a purely epiphytic parasite, but also it is able to select within the leaf of the host those regions better supplied with food and water than the loose parenchyma. If the fungus is stimulated to place haustoria in cells joining the bundles to the palisade layer, or if haustoria enter the

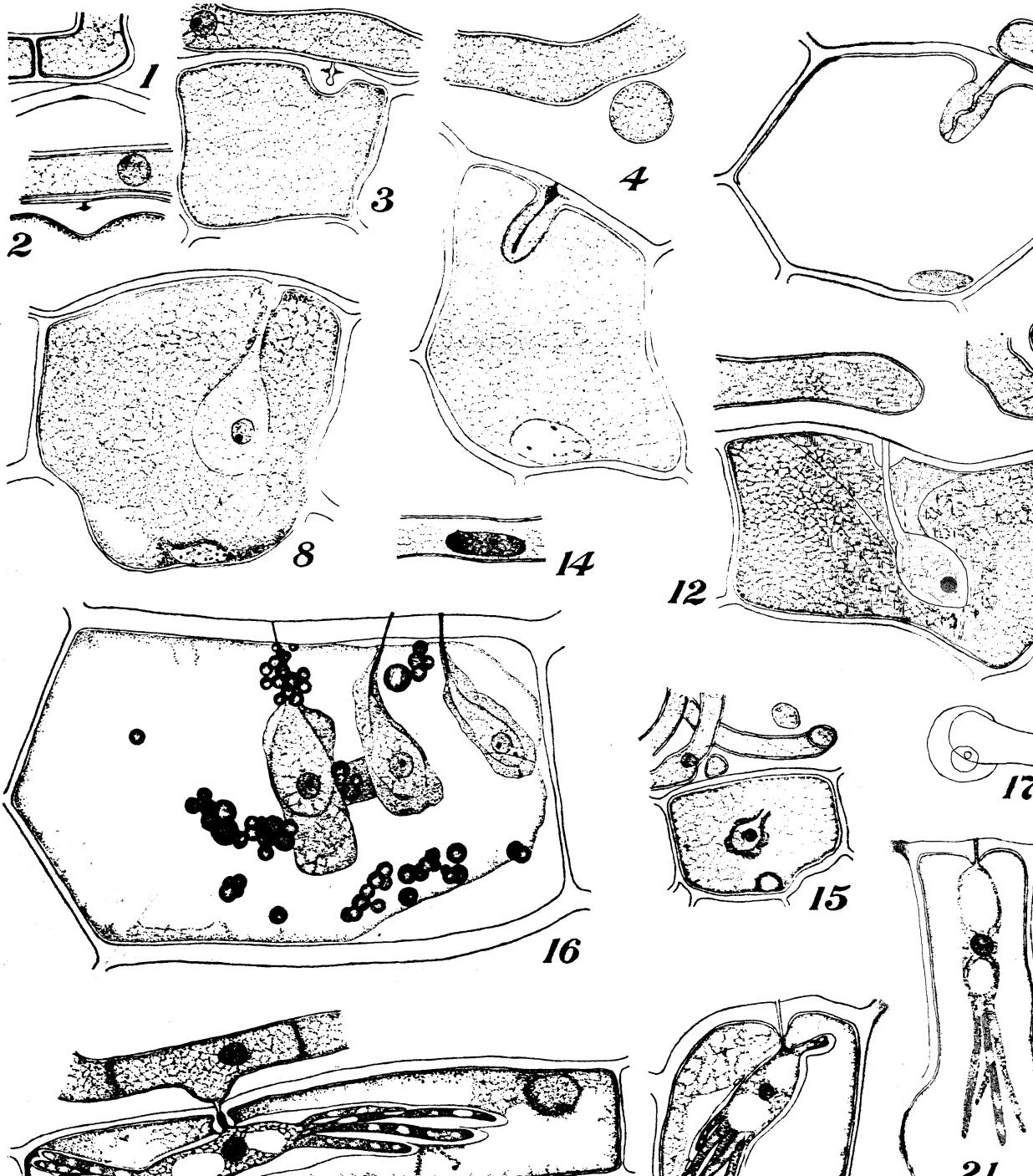
palisade cells, they are still at the very source of that supply of food by which the plant maintains its vigor and activity. The proportion of cases to be found in which the intercellular hyphæ do develop haustoria in these more favorable regions is larger than casual observation would lead one to think. By a study of the cells of the host-leaf in sections preceding and following the section which contains the haustorium, it may often be determined that the fungus actually has placed its absorbing organs in one of the favorable cells. It should not be thought, however, that this is done invariably. In Xanthoxylum the leaves are so constructed that the bundles are near the lower epidermis through the stomata of which the fungus finds access. All of the mesophyll cells, therefore, have a surplus of food. This fact would account for the number of mesophyll cells which contain haustoria.

If it be demonstrated that the mildew on Xanthoxylum does not possess typical haustoria under any conditions, it will be important to discover whether the apparently stunted haustoria can absorb food for the fungus as well as typical organs. If they are not effective, how far is the fungus nourished by saprophytic methods? In the first place, the intercellular spaces of the leaves of this particular host are abundantly supplied with organic material. On the part of the parasite itself it is noticeable that, when under the stomata, the intercellular hyphæ encounter parenchyma cells which block the way, the cells of the hyphæ are short, thick, and vigorous; not only when the haustoria are produced near, but also when the hyphæ have haustoria at some distance from the stomata. But the hyphæ which do not encounter such cells, but run a comparatively uninterrupted course before constructing haustoria, are more or less attenuated and are less vigorous in appearance. If some cell of the hypha in such a case lies in contact with a parenchyma cell (*fig. 33*, penultimate cell), that cell of the hypha is short, thick, and vigorous. Büsgen's results would seem to show that Erysiphe does not succeed as a saprophyte. At least, he found that its germinating tubes had a limited growth under the conditions

he was able to establish. He did not succeed in infecting pieces of leaves arranged for the purpose. If, under natural conditions of infection, the germinating tubes have a limited growth unless supplied with nutrient material from the host in some way, it is not clear how they can obtain it so as to grow down into the leaves for comparatively long distances, as Phyllactinia does, before producing haustoria, unless the fungus appropriates intercellular material. The subject is still open for investigation. It is possible, of course, that the first side branch, passing into the interior of the leaf, places an absorbing organ near the stoma and thus supports the whole branching system until other side branches in other stomata are able to assist.

There is some evidence to support the possibility of intercellular nutrition in this fungus, as has been established for others by Nordhausen, Ward, Büsgen, and Frank. This investigation, however, has not embraced the question of how far, if at all, the abundant intercellular material in Xanthoxylum leaves is available, though some facts were observed which suggest the possibility of intercellular nutrition of the parasite which have not been observed in other hosts infested by Phyllactinia.

There are two points connected with my observation of Erysiphe on the geranium leaf which may be mentioned briefly. At the end of a section where the scissors with which the pieces of leaves were cut had destroyed the section in part, a haustorium was found in what was clearly a subepidermal cell. Its connection with the surface was destroyed, but there is no doubt that it was a normal haustorium in one of these cells. Whether it was due to the chance presence of Phyllactinia on this host, whether Erysiphe occasionally adopts the practice of *Uncinula Salicis*, or whether it was Uncinula itself on this host, could not be determined. With this circumstance is related a note on page 9 of Ellis and Everhart's *Pyrenomycetes*. This note corrected the mistake of the artist (F. W. Anderson) who, it was supposed, wrongly represented (*fig. 3, pl. 1*) a germinating tube from a conidium of *Sphaerotheca Castagnei* as entering a stoma. My observations render it probable that the artist saw what he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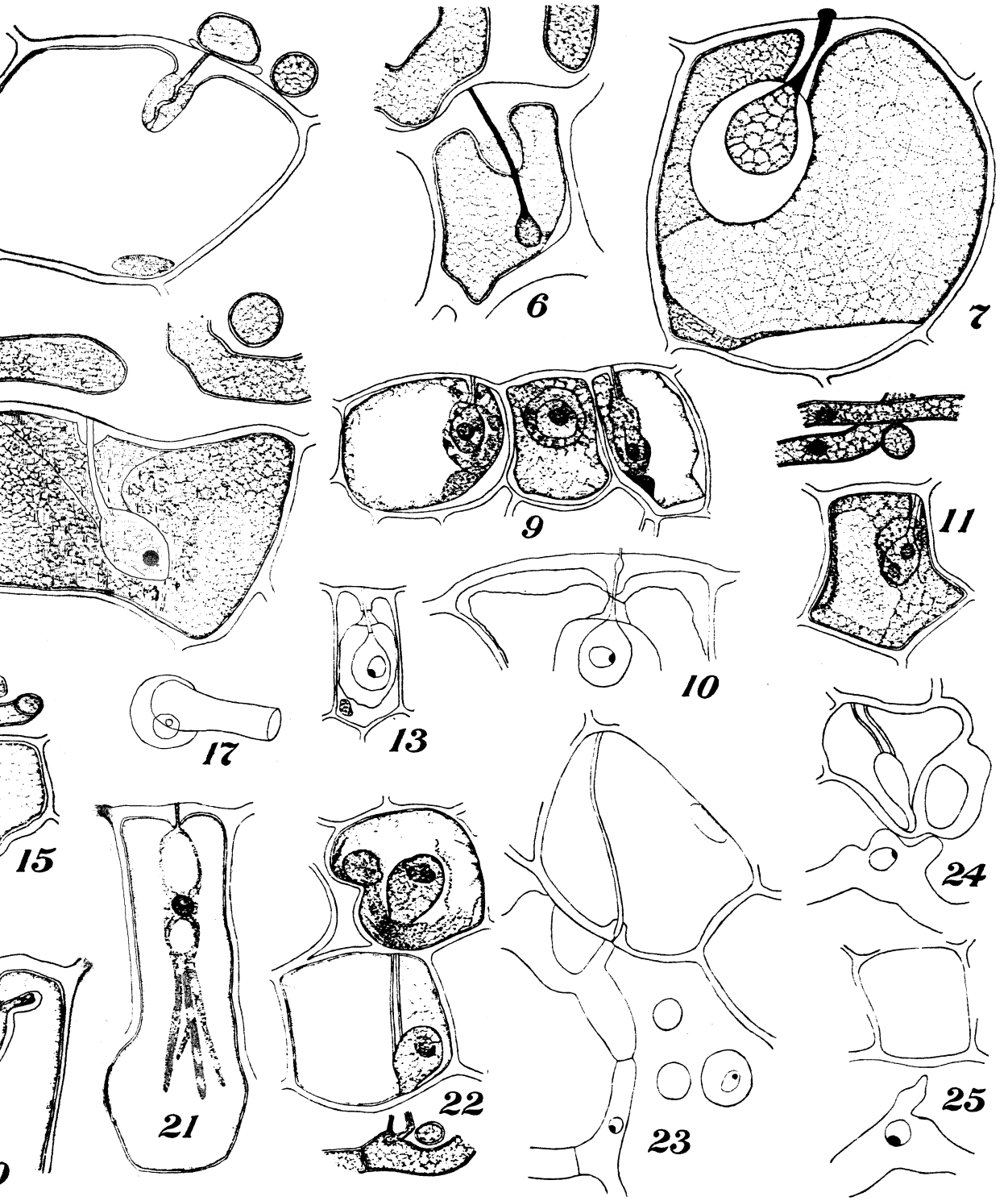
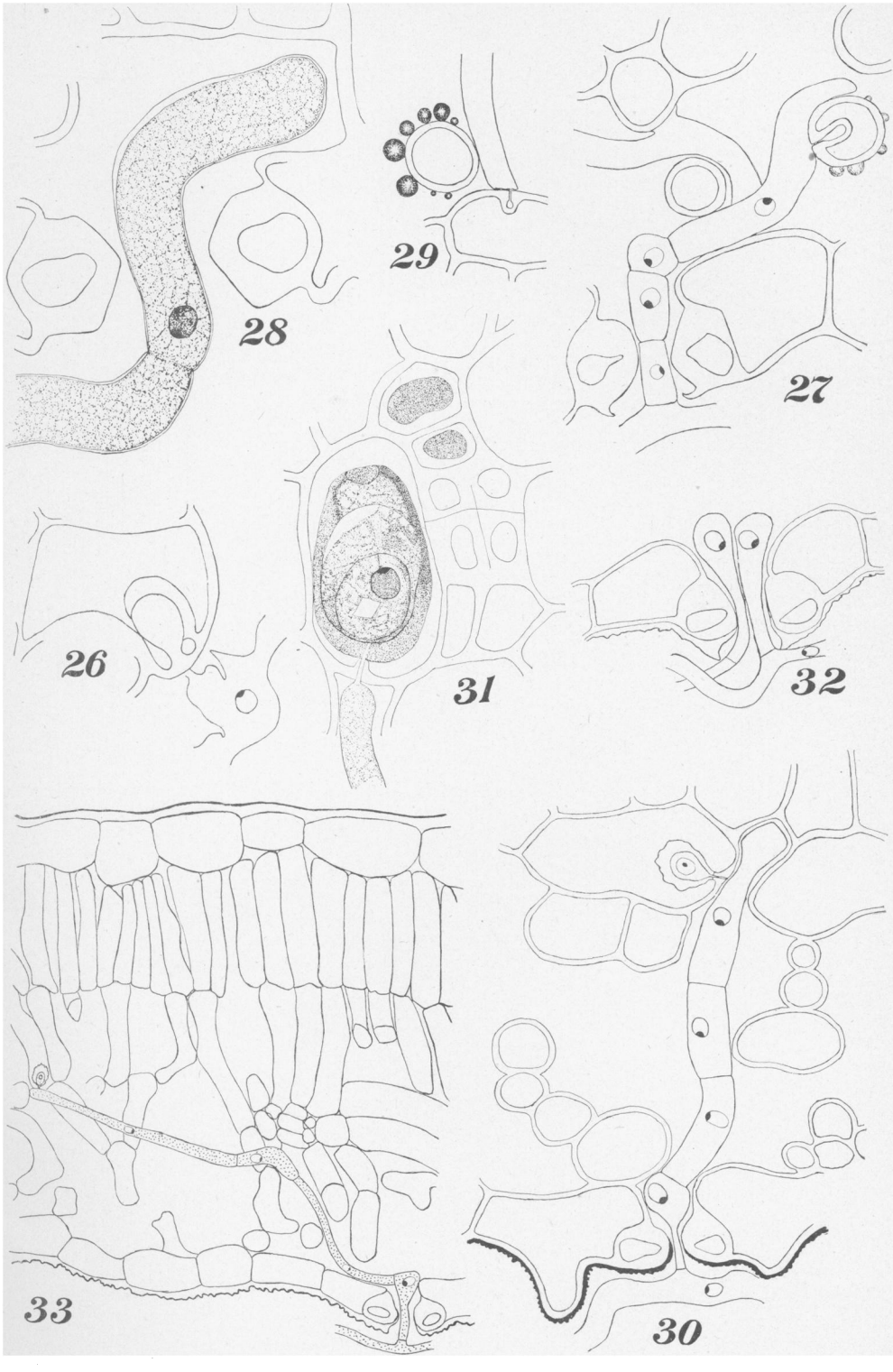


on HAUSTORIA OF ERYSIPHEÆ



SMITH on HAUSTORIA OF ERYSIPHEÆ

represented, though it may have been a tube of Phyllactinia. Whether or not Sphærotheca has a similar habit to Uncinula, I have not determined.

It is a pleasure to acknowledge the assistance I have had in this investigation from Professor C. R. Barnes, under whom the work was begun, and from Professor R. A. Harper, under whom it was mainly accomplished.

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LIST OF GENERA AND SPECIES STUDIED.

Erysiphe communis (Wallr.) on *Geranium maculatum* L.
——— *graminis* DC. on *Poa pratensis* L.
——— *Galeopsidis* DC. on *Scutellaria lateriflora* L.
——— *Galeopsidis* DC. on *Galeopsis Tetrahit* L.
——— *communis* on *Polygonum aviculare* L.
——— *Cichoracearum* DC. on *Eupatorium perfoliatum* L.
Sphærotheca Castagnei Lév. on *Bidens cernua* L.
Podosphæra Oxycanthæ DC. on *Prunus* sp.
Microsphæra Russellii Clinton on *Oxalis corniculata stricta* Sav.
Uncinula Salicis DC. Wint. on *Salix discolor* Muhl.
——— *necator* Schw. on *Vitis* sp.?
——— *Clintonii* Peck, on *Tilia Americana* L.
Phyllactinia suffulta (Reb.)[1] on *Fraxinus pubescens* Lam.; *Cratægus punctata* Jacq.; *Xanthoxylum Americana* Mill; *Corylus Americanum* Walt; *Cornus stolonifera* Michx.

Such forms as are not discussed in the preceding pages have not been found to differ so materially from *Erysiphe communis* as to require a separate discussion of their haustoria.

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[1] The material has not been examined with reference to the classification of Phyllactinia proposed by Palla (**24**, p. 65).

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EXPLANATION OF PLATES XI AND XII.

All the figures were drawn by the aid of the Abbé camera.

Magnification of *figs. 9, 11, 13, 22 to 30*, 900 diameters; *figs. 16, 28*, 2000 diameters; *figs. 32, 33*, 400 diameters; all other figures, 1800 diameters.

FIGS. 1–15. *Erysiphe communis* on *Geranium maculatum.*

FIG. 1. First stage in penetration, marked by spot on inner surface of host wall.

FIG. 2. Penetrating tube just beginning to make its way through epidermal wall. Wall thickens as tube grows.

FIG. 3. Having enlarged at a point in line with the inner boundary of the epidermal wall the tube continues its growth.

FIG. 4. Further stage in penetration. Cellulose papilla elongates as the tube grows. Distal end of cellulose in growth begins to show signs of disintegration. Nucleus of host-cell on basal wall of cell.

FIG. 5. Distal end of penetrating tube now begins to enlarge. Cellulose further disorganized. The proximal end of ingrowth not disintegrating, but destined to remain as collar about neck of haustorium.

FIG. 6. Young haustorium escapes from cellulose thickening and is developing without sheath.

FIG. 7. Stage before entrance of haustorium nucleus. Stretched and intruded plasmic membrane of host-cell forms bounding membrane of sheath, which in this case is devoid of contents.

FIG. 8. Haustorium without sheath.

FIG. 9. Middle haustorium in optical section, the two at end in longitudinal section. Sheath present about haustoria.

FIG. 10. Plasmolyzed cell showing collar as an ingrowth of cellulose from cell wall. Partly in perspective.

FIG. 11. Haustorium with two nuclei. Septum also found across neck.

FIG. 12. Haustorium without sheath. Protoplasm of host-cell adherent to sides of neck.

FIG. 13. Haustorium shows septum across neck.

FIG. 14. Elongated nucleus in mycelial hypha.

FIG. 15. Membrane absent from sheath, which here consists of lumps of disintegrated cellulose adhering to body of haustorium.

FIGS. 16–17. *Erysiphe Cichoracearum* on *Eupatorium perfoliatum.*

FIG. 16. Basal cell of hair containing haustoria.

FIG. 17. Shows unstained area about point of penetration. Remainder of host wall staining darkly.

FIGS. 19–21. *Erysiphe graminis* on Poa. Three branching haustoria.

FIGS. 22–26. *Uncinula Salicis* on *Salix discolor.*

FIG. 22. Epidermal cell containing one haustorium, while from the same appressorium another penetrating tube passes through the epidermal cell into the subepidermal cell. The sheaths are not visible.

FIGS. 23–26. Various forms of appressoria giving rise to two or more haustoria.

FIG. 27. Modified haustorium arising from appressorium and penetrating mesophyll cell. Host-cell shows exudations on its exterior. A second appressorium also shown.

FIG. 28–33. *Phyllactinia suffulta; figs. 28, 31* on *Fraxinus pubescens; fig. 30* on *Cornus* sp.; *figs. 27, 29, 32, 33* on *Xanthoxylum Americanum.*

FIG. 28. Part of an intercellular hypha. Guard cells drawn apart and hypha not compressed, as in *figs. 32, 33.*

FIG. 29. Smaller haustorium, and arising from end of hypha. Exudation on cell of leaf shown.

FIG. 30. Intercellular hypha consisting of four cells. Haustorium arising from side of distal cell. Host-cell below palisade cells.

FIG. 31. Haustorium arising from end of intercellular hypha and penetrating parenchyma cell connected with bundle. Haustorium contains a crystal.

FIG. 32. Stoma containing initial cells of two intercellular hyphæ, one a side branch. Origin of the other is indeterminate.

FIG. 33. Intercellular hypha, with haustorium in cell joining bundle to palisade cell. Middle cell of hypha is short and thick, and lies on cells of host.